

Systematic detection and classification of cultivable microbes present in the Spacecraft Assembly Facility (SAF) at the Jet Propulsion Laboratory (JPL) were carried out using classical microbial phylogeny and advanced molecular microbial ecology methods. This work is being carried out to understand the microbial diversity in a SAF environment that could potentially contaminate an assembled spacecraft. Contamination of spacecraft surfaces with terrestrial microbes would compromise the interpretation of results from in-situ life detection studies or sample return missions. Fifty samples were collected from various locations of a JPL-SAF whose air circulation and humidity were controlled and maintained to the cleanliness of a class 100K clean-room. Sampling locations include both unclassified (entrance floors, shoe-cleaner, air-shower, ante-room, and air-lock), and classified (clean-room floors, clean-room tables and cabinet tops) areas. All samples were analyzed for the incidence of aerobic spore-formers (using Tryptic Soy Agar) and for total aerobic heterotrophs (using R2 Agar). Spore-former incidence ranged from 0 to 3×10^{-1} CFU per 25cm^2 in the unclassified area whereas only 2 out of 25 samples collected in the unclassified areas contained spore-formers. However, the counts of total heterotrophs were about 100 times higher when compared to the spore-formers in the unclassified area and 9 out of 25 samples collected in the classified area contained a range of heterotrophs from 5×10^1 to 4×10^2 cfu/ 25cm^2 . Representatives of the spore-formers (31 strains) and total heterotrophs (40 strains) were identified by 16S rDNA sequence analysis to determine their phylogenetic relatedness. The spore-formers clustered to 8 known *Bacillus* and *Paenibacillus* species and 6 strains were identified as novel species of the genera *Bacillus* (2), *Paenibacillus* (2), *Ureibacillus* (1), and one new genus. Among the forty heterotrophs, 5 clusters were tightly affiliated with genera, such as *Bacillus* (5), *Staphylococcus* (2), and members of the families actinomycetales (3), streptomyces (1), and micrococcoidaceae (10). However, 19 of the strains isolated clustered to a very distinct clade and formed a relatively close association with the Cytophaga-Flavobacteria-Bacteroides-Taxebacter (CFBT) group. The physiological novelties of these species such as resistance to hydrogen peroxide, desiccation, etc. will be presented. Isolation of microbes that are resistant to hydrogen peroxide has significant implications in the assembly of spacecraft because vapor hydrogen peroxide is the low-heat sterilization technology of choice for spacecraft hardware.